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L23	L22 AND equilibrium	918	L23	
L22	L21 AND density	4961	L22	
L21	L20 AND gradient	7337	L21	
L20	vesicle	19333	L20	
L19	Berg-Eric-A.IN.	. 5	L19	
L18	Fine-Richard-E.IN.	2	L18	
L17	dense core vesicles	54	L17	
DB=USF	PT,PGPB; PLUR=YES; OP=ADJ			
L16	L14 AND vesicle	35	L16	
L15	L14 AND microsome	2	L15	
L14	L13 AND sucrose gradient	206	L14	
L13	((435/7.1 435/317.1)!.CCLS.)	6316	L13	
DB=USF	PT,PGPB,JPAB,EPAB,DWPI; PLUR=YES; OP=ADJ			
L12	L11 AND equilibrium gradient	32	L12	
L11	sucrose gradient	2910	L11	
DB=USF	PT,PGPB; PLUR=YES; OP=ADJ			
L10	L9 AND equilibrium density gradient	1	L10	
L9	L8 AND sucrose gradient	265	L9	
L8	((435/325 435/352 435/366 435/368)!.CCLS.)	11207	L8	
L7	L6 AND equilibrium density gradient	4	L7	
L6	L3 AND sucrose gradient	482	L6	
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· L4.	L3 sucrose velocity size gradient	Ö	L4	
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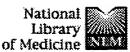
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<u>L12</u>	L11 AND equilibrium	75	<u>L12</u>	
<u>L11</u>	L10 AND vesicle	361	<u>L11</u>	
<u>L10</u>	sucrose gradient	2917	<u>L10</u>	
<u>L9</u>	L8 AND velocity	61	<u>L9</u>	
<u>L8</u>	L7 AND equilibrium	402	<u>L8</u>	
<u>L7</u>	sucrose gradient	2917	<u>L7</u>	
<u>L6</u>	L5 AND sucrose gradient	5	<u>L6</u>	
<u>L5</u>	microsome preparation	85	<u>L5</u>	
<u>L4</u>	sucrose equilibrium density gradient	7	<u>L4</u>	
<u>L3</u>	sucrose velocity size gradient	1	<u>L3</u>	
<u>L2</u>	velocity size AND equilibrium density	1	<u>L2</u>	
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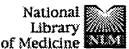
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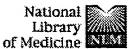
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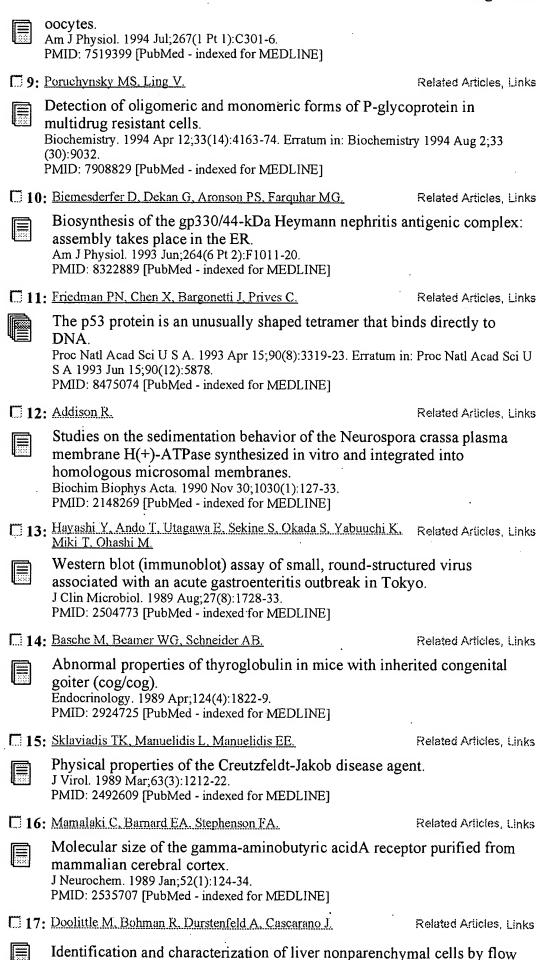


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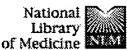
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Purification of synaptic vesicles from elasmobranch eleuse of biophysical criteria to demonstrate purity. Biochemistry. 1978 Apr 4;17(7):1188-99. PMID: 418798 [PubMed - indexed for MEDLINE]	ectric organ and the
17: Zakar T, Toth M.	Related Articles, Links
Characterization of the cytoplasmic androgen recentor	of rat seminal

vesicle.

Steroids. 1977 Dec;30(6):751-64.

PMID: 611640 [PubMed - indexed for MEDLINE]

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=> s dense core vesicles AND equilibrium density gradient AND size gradient

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L2 2 DENSE CORE VESICLES AND EQUILIBRIUM DENSITY GRADIENT AND SIZE GRADIENT

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METHOD OF PURIFICATION OF NEUROPEPTIDE CONTAINING VESICLES FROM THE BRAIN; ANALYZING DENSE CORTESSICLES FROM BRAIN TISSUES; OF IN BRACELLS, INCUBATE WITH EXTRACTION BUFFER, MONITOR VESSICLE TISSUES
                                                                                           IN BRAIN
IN
        Berg Eric A; Fine Richard E
PA
        Boston University (1308)
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        US 2002090655
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                                   20011031
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        Utility; Patent Application - First Publication
FS
        CHEMICAL
        APPLICATION
CLMN
        15
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         6 Figure(s).
       FIGS. IA and 1B disclose the separation of substance Pcontaining vesicles
        in a sucrose velocity gradient. Preparations of microsomes from ON and
        LGN/SC respectively, were centrifuged in a 10-50% sucrose velocity gradient and analyzed for substance P by RIA. A, Substance P distribution of fractionated ON microsomes; and B, Substance P distribution of fractionated LGN/SC microsomes by radioimmunoassay.
       FIGS. 2A-2E disclose the fractionation of substance P-containing vesicles
        in a sucrose equilibrium gradient. Fractions containing substance P from
        FIG. 1 were pooled, concentrated, and then loaded on an equilibrium
        density sucrose gradient (2550%). Fractions were analyzed for A
        substance P content (ON); B, substance P content (LGN/SC) by RIA; C,
        individual synaptic vesicle membrane proteins (SV2, synaptotagmin, and
        synaptophysin) (ON); D, individual synaptic vesicle membrane proteins
      (SV2, synaptotagmin, synaptotagmin, synaptophysin, and synaptobrevin) (LGN/SC); E, synaptotagmin IV (LGN/SC) by Western blot. FIGS. 3A-3E disclose co-sedimentation of vesicle-associated proteins with
        substance P inLGN/SC microsomes fractionated by size and density.
        Fractions containing substance P from FIG. 2 were analyzed for A,
        secretogranin II; B, beta APP (C8); C, Rab3, D, alpha-synuclein; E, BDNF
        by Western blot.
       FIG. 4 discloses immunoadsorption of synaptic vesicle membrane proteins
        from substance P-containing fractions. Fractions containing substance P
        from FIG. 2 were immunoadsorbed with mouse IgG or synaptophysin Ab linked
        magnetic beads (2 or 4 mgs) (Dynal) as per the manufacturers instructions. Samples were analyzed for specific synaptic vesicle
        membrane proteins (SV2, synaptotagmin, synaptophysin, and synaptobrevin)
        by Western blot.
      FIGS. 5A and 5B disclose electron micrographs of an immunolabeled, negatively stained DCV preparation from LGN/SC. Fractions containing substance P from an equilibrium gradient were fixed, adhered to formvar,
        carbon coated nickel grids and A, immunolabeled with mouse IgG and
        colloidal gold conjugated antibodies (12 nm); or B, immunolabeled with synaptophysin and colloidal gold conjugated antibodies (12 nm).
       FIGS. 6A-6D disclose electron micrographs of immunolabeled thin sections
        from a DCV preparation from LGN/SC. Fractions containing substance P from
        an equilibrium gradient were fixed, embedded, sectioned and immunolabeled with A, rabbit IgG an colloidal gold conjugated synaptotagmin and colloidal gold conjugated antibodies (12 nm); and D, BDNF and colloidal
        gold conjugated antibodies (6 nm).
=> s velocity size gradient AND equilibrium density gradient
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                 2 VELOCITY SIZE GRADIENT AND EQUILIBRIUM DENSITY GRADIENT
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ANSWER 1 OF 1 IFIPAT COPY HT 2003 IFI
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METHOD OF PURIFICATION OF NEUROPEPTIDE CONTAINING VESICLES FROM THE
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         BRAIN; ANALYZING DENSE CORE VESSICLES FROM BRAIN TISSUES; OBTAIN BRAIN
         CELLS, INCUBATE WITH EXTRACTION BUFFER, MONITOR VESSICLE TISSUES
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 DT
         Utility; Patent Application - First Publication
FS
         CHEMICAL
         APPLICATION
 CLMN
         15
 GΙ
          6 Figure(s)
        FIGS. 1A and 1B disclose the separation of substance Pcontaining vesicles
         in a sucrose velocity gradient. Preparations of microsomes from ON and
         LGN/SC respectively, were centrifuged in a 10-50% sucrose velocity
         gradient and analyzed for substance P by RIA. A, Substance P distribution of fractionated ON microsomes; and B, Substance P distribution of fractionated LGN/SC microsomes by radioimmunoassay.
        FIGS. 2A-2E disclose the fractionation of substance P-containing vesicles
         in a sucrose equilibrium gradient. Fractions containing substance P from
         FIG. 1 were pooled, concentrated, and then loaded on an equilibrium density sucrose gradient (2550%). Fractions were analyzed for A, substance P content (ON); B, substance P content (LGN/SC) by RIA; C, individual synaptic vesicle membrane proteins (SV2, synaptotagmin, and synaptophysin) (ON); D, individual synaptic vesicle membrane proteins
         (SV2, synaptotagmin, synaptotagmin, synaptophysin, and synaptobrevin) (LGN/SC); E, synaptotagmin IV (LGN/SC) by Western blot.
        FIGS. 3A-3E disclose co-sedimentation of vesicle-associated proteins with
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         by Western blot.
        FIG. 4 discloses immunoadsorption of synaptic vesicle membrane proteins
         from substance P-containing fractions. Fractions containing substance P
         from FIG. 2 were immunoadsorbed with mouse IgG or synaptophysin Ab linked magnetic beads (2 or 4 mgs) (Dynal) as per the manufacturers instructions. Samples were analyzed for specific synaptic vesicle
         membrane proteins (SV2, synaptotagmin, synaptophysin, and synaptobrevin)
         by Western blot.
        FIGS. 5A and 5B disclose electron micrographs of an immunolabeled,
         negatively stained DCV preparation from LGN/SC. Fractions containing
         substance P from an equilibrium gradient were fixed, adhered to formvar,
         carbon coated nickel grids and A, immunolabeled with mouse IgG and
        colloidal gold conjugated antibodies (12 nm); or B, immunolabeled with synaptophysin and colloidal gold conjugated antibodies (12 nm).

FIGS. 6A-6D disclose electron micrographs of immunolabeled thin sections
         from a DCV preparation from LGN/SC. Fractions containing substance P from an equilibrium gradient were fixed, embedded, sectioned and immunolabeled with A, rabbit IgG an colloidal gold conjugated synaptotagmin and
         colloidal gold conjugated antibodies (12 nm); and D, BDNF and colloidal
         gold conjugated antibodies (6 nm):
 => s size gradient AND equilibrium gradient AND vesicle
    26 FILES SEARCHED...
    48 FILES SEARCHED...
                  3 SIZE GRADIENT AND EQUILIBRIUM GRADIENT AND VESICLE
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10147014 IFIPAT;IFIUDB;IFICDB
METHOD OF PURIFICATION OF NEUROPEPTIDE CONTAINING ***VESICLES***

DUPLICATE 1

FROM

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       Boston University (1308)
PA
       us 2002090655
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US 2000-244971P
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                                20001101 (Provisional)
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       Utility; Patent Application - First Publication
DT
FS
       CHEMICAL
       APPLICATION
CLMN
       15
        6 Figure(s).
GI
      FIGS. IA and 1B disclose the separation of substance Pcontaining
         ***vesicles*** in a sucrose velocity_gradient. Preparations of
       microsomes from ON and LGN/SC respectively, were centrifuged in a 10-50%
       sucrose velocity gradient and analyzed for substance P by RIA. A, Substance P distribution of fractionated ON microsomes; and B, Substance
       P distribution of fractionated LGN/SC microsomes by radioimmunoassay.
      FIGS. 2A-2E disclose the fractionation of substance P-containing
          ***vesicles*** in a sucrose ***equilibrium***
                                                                             ***gradient***
       Fractions containing substance P from FIG. 1 were pooled, concentrated,
       and then loaded on an equilibrium density sucrose gradient (2550%).
       Fractions were analyzed for A, substance P content (ON); B, substance P content (LGN/SC) by RIA; C, individual synaptic ***vesicle***
       membrane proteins (SV2, synaptotagmin, and synaptophysin) (ON); D, individual synaptic ***vesicle*** membrane proteins (SV2,
       synaptotagmiń, synaptotagmin, synaptophysin, and synaptobrevin) (LGN/SC);
E, synaptotagmin IV (LGN/SC) by Western blot.
                                                           ***vesicle*** -associated
      FIGS. 3A-3E disclose co-sedimentation of
       proteins with substance P inLGN/SC microsomes fractionated by size and
       density. Fractions containing substance P from FIG. 2 were analyzed for A, secretogranin II; B, beta APP (C8); C, Rab3, D, alpha-synuclein; E,
       BDNF by Western blot.
      FIG. 4 discloses immunoadsorption of synaptic
                                                                 ***vesicle***
       proteins from substance P-containing fractions. Fractions containing
       substance P from FIG. 2 were immunoadsorbed with mouse IgG or
       synaptophysin Ab linked magnetic beads (2 or 4 mgs) (Dynal) as per the
       manufacturers instructions. Samples were analyzed for specific synaptic
          ***vesicle*** membrane proteins (SV2, synaptotagmin, synaptophysin, and
       synaptobrevin) by Western blot.
      FIGS. 5A and 5B disclose electron micrographs of an immunolabeled, negatively stained DCV preparation from LGN/SC. Fractions containing substance P from an ***equilibrium**** ***gradient*** were fixed,
       adhered to formvar, carbon coated nickel grids and A, immunolabeled with
       mouse IgG and colloidal gold conjugated antibodies (12 nm); or B,
       immunolabeled with synaptophysin and colloidal gold conjugated antibodies
      FIGS. 6A-6D disclose electron micrographs of immunolabeled thin sections
       from a DCV preparation from LGN/SC. Fractions containing substance P from an ***equilibrium*** ***gradient*** were fixed, embedded,
                                                             were fixed, embedded,
       sectioned and immunolabeled with A, rabbit IgG an colloidal gold conjugated synaptotagmin and colloidal gold conjugated antibodies (12
       nm); and D, BDNF and colloidal gold conjugated antibodies (6 nm).
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      2002-435699 [46]
AN
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      N2002-342949
                             DNC C2002-123804
DNN
      determining contents of purified dense core ***vesicles***, comprises extracted brain samples which are purified with
      extracted brain samples which are purified with predetermined fold.
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WO 2002037104 A2 WO 2001-US45485 20011031; US 2002090655 A1 Provisional US
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FDT AU 2002020071 A Based on WO 0237104

PRAI US 2000-244971P 20001101; 05 2001-1898 20011031

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ICS A61K035-30; C12N001-00; G01N033-567

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